Configurational Changes of Retinal in the Triplet State: Picosecond Time-Resolved Absorption Spectroscopy on the 7-Cis, 11-Cis, and 13-Cis Isomers and High-Performance Liquid Chromatography Analysis of Photoisomerization

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The triplet state of retinal was produced from the 7-cis, 11-cis, and 13-cis isomers by direct excitation with 355-nm ~10-ps pulses in deoxygenated *n*-hexane solution at room temperature. A set of transient absorption spectra was recorded for each isomer, in the time domain within 5 ns after excitation. The results indicated the following: (1) The 7-cis and 11-cis isomers initially produce their own short-lived, primary "7-cis" and "11-cis" triplet species which relax into the common, stationary "all-trans" triplet species. (2) The 13-cis isomer produces two different stationary triplet species, i.e., one its own "13-cis" and the other the above "all-trans" triplet species. (3) The above cis isomers produce also "all-trans" triplet species immediately after excitation. The presence of two different long-lived, stationary triplet species, which were revealed by a previous study by resonance Raman spectroscopy, is confirmed, and short-lived, primary triplet species produced from the 7-cis and 11-cis isomers have been identified, in addition to the one from the 9-cis isomer, which was identified in a previous transient absorption study. The products of isomerization by benzil-sensitized triplet excitation of the all-trans, 7-cis, 9-cis, 11-cis, and 13-cis isomers were analyzed by means of high-performance liquid chromatography. The major product of isomerization from the cis isomers was the all-trans isomer. On the basis of the above results, the mechanisms of isomerization via the triplet state are discussed.

The structural changes of the retinylidene chromophore of rhodopsin and bacteriorhodopsin are triggered by the excitation of the chromophore as a result of the absorption of light. These changes are due to the fact that the stable configurations in the excited state are different from those in the ground state. It is of interest that particular configurational changes are selected by nature to carry out the physiological functions of the rhodopsins. The retinylidene chromophore in rhodopsin is originally in the 11-cis configuration, and it is transformed into the all-trans configuration after the absorption of light.[1] By contrast, the retinylidene chromophore in bacteriorhodopsin is believed to be initially in the all-trans configuration, and then it is transformed into the 13-cis configuration.[2] Therefore, it is most important to address the question of what are the stable configurations in the excited states, since little is known about these configurations. As a first step in a series of investigations into the excited-state configurations, as well as into the molecular dynamics of excitation and subsequent relaxation of the retinylidene chromophore, we have examined the configurations of free retinal in the triplet state, which are produced from different cis–trans isomers.

In a previous transient Raman investigation,[3] we found two triplet species with different configurations, i.e., "all-trans" and "13-cis", which were supposed to relax into the ground state as the all-trans and 13-cis isomers, respectively. Resonance Raman spectroscopy is a sensitive tool for the detection of configurational differences and provides more definitive structural information than does electronic absorption spectroscopy. However, transient absorption spectroscopy is much more sensitive in detecting short-lived species in their excited states. In addition, Raman intensity is strongly dependent on the resonance conditions. Therefore, a set of spectral data of the $T_n \leftarrow T_1$ absorption for all the isomers is absolutely necessary if we are to be able to draw any conclusions about the different kinds of triplet configurations from the comparison of resonance Raman spectra.

The $T_n \leftarrow T_1$ absorption data of isomeric retinal have been recorded under a variety of different conditions; the definition of conditions includes the method of excitation (direct or sensitized), the time of delay after excitation, the solvent, and the temperature. Similar triplet spectra have been reported for the all-trans and 11-cis isomers (100 ns after direct excitation at 337 nm)[4] and also for the all-trans, 9-cis, 11-cis, and 13-cis isomers immediately after excitation at 265 nm,[5] in *n*-hexane solution at room temperature in both cases. Indistinguishable spectra have been recorded, by both direct (delay 4–20 μs, wavelength not specified) and anthracene-sensitized excitation, for the all-trans, 7-cis, 7,9-cis,cis, and 11-cis isomers in benzene solution at room temperature.[6] However, Veyret et al.[7] reported different triplet spectra (direct excitation at 347 nm, delay not specified) for the all-trans and 11-cis isomers, in *n*-hexane solution at room temperature. They concluded that the transient absorption of the 11-cis isomer is due to the triplet state of this isomer as well as to the triplet state of the all-trans isomer.

The $T_n \leftarrow T_1$ absorption spectra in polar solvents are completely different from those in nonpolar solvents.[8] A set of spectra, which were different from one another, was recorded for the all-trans, 9-cis, 11-cis, and 13-cis isomers (immediately and 18 μs after direct excitation at 265 nm) in methanol at room temperature.[9] Different spectra were also obtained in the case of EPA glass, at liquid N_2 temperature for the all-trans, 7-cis, 11-cis, and 13-cis isomers (8–10 μs after direct excitation at 350–400 nm).[10]

A complete set of electronic absorption data for the triplet state, to be used for the interpretation of the above results of resonance Raman spectroscopy, is not available, and some of the results in the literature are contradictory. Recently, we reported a pair of transient absorption spectra for the all-trans and 9-cis isomers of retinal, which indicated a configurational change of the 9-cis isomer in the triplet state.[11] In the present investigation, we have

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attempted to record the transient absorption spectra of the 7-cis, 11-cis, and 13-cis isomers, under the conditions of our previous resonance Raman and transient absorption measurements, i.e., by direct excitation at 355 nm in *n*-hexane solution at room temperature.

The products of isomerization via the triplet state should provide indirect information about the configuration of the triplet species just before relaxation. However, under the present experimental conditions, i.e., direct $S_n \leftarrow S_1$ excitation, retinal can isomerize via either the singlet or the triplet state. It is necessary to estimate the isomerization yield strictly via the triplet state. The quantum yield of sensitized photoisomerization for the all-trans and 11-cis isomers[4] have been reported for *n*-hexane solutions. However, the above data were not sufficient for the interpretation of the transient absorption data for the complete set of the cis–trans isomers. Therefore, we analyzed the products of sensitized photoisomerization by using benzil as a sensitizer, for the all-trans, 7-cis, 9-cis, 11-cis, and 13-cis isomers.

We have confirmed the presence of two different stationary triplet species that we found in a previous resonance Raman study, and have newly found additional two different short-lived, triplet species. In addition, we obtained evidence for isomerization through configurational changes in the triplet state.

Experimental Section

The 7-cis, 11-cis, and 13-cis isomers of retinal were isolated by the method described previously.[12] The solvent *n*-hexane (HPLC grade) was purchased from Wako Chemical Co. Inc. and dried over molecular sieves. A solution of each isomer in hexane ($(3.6–7.6) \times 10^{-5}$ M; $abs_{355\,nm} = 2.0$) was deoxygenated by bubbling N_2 through it and then sealed in a 1-cm cell. The triplet state was produced by exciting the sample with a 355-nm picosecond pulse (20–25-ps duration) and probed with a picosecond continuum of the same pulse duration, which was produced by focusing the 1.06-μm pulse on a cell that contained D_2O. The delay time of the probe pulse from the pump pulse was from "0 ps" (the pump and the probe pulses are temporally overlapped) to 5 ns. The system for recording transient absorption has already been reported.[13] Each spectrum in the figures, i.e., the one for each sample solution at one delay time, is the result of accumulating four observed spectral data without smoothing. The solution was replaced by fresh solution after each measurement. This procedure maintained the sum of all the isomerization products below 6% of the total dissolved material.

In sensitized photoisomerization, 1×10^{-4} M isomeric retinal and 1×10^{-2} M benzil were dissolved into *n*-hexane, and the solution was deoxygenated and then irradiated with yellow light (220 W) above 450 nm (filtered through water and a Toshiba V-Y47 filter) for 2 min. The irradiated solutions were analyzed by conventional high-performance liquid chromatography (HPLC).[12] The amounts of isomerization products were estimated from the area under the peak for each isomer in the elution profile, with a correction factor for the absorption of each isomer at the detection wavelength of 360 nm.

Results and Discussion

Time-Resolved Absorption Spectra within 100 ps after Excitation. Figure 1 shows the configurations of the isomers of retinal in the ground state, i.e., (a) all-trans, (b) 7-cis, (c) 9-cis, (d) 11-cis, and (e) 13-cis. Depending on the structure of the cis group, the cis configurations are classified into unmethylated-cis (7-cis and 11-cis) and methylated-cis (9-cis and 13-cis) configurations. Depending on the location of the cis bend, they are also classified into terminal-bent (7-cis and 13-cis) and central-bent (9-cis and 11-cis) configurations. These starting ground-state configurations should affect the structures of triplet species and their molecular

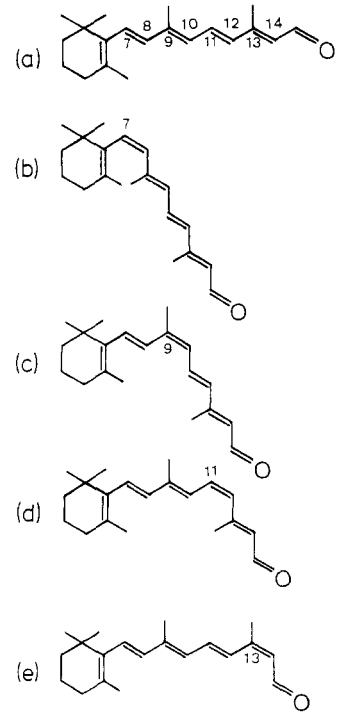

Figure 1. Configurations of retinal isomers in the ground state: (a) all-trans, (b) 7-cis, (c) 9-cis, (d) 11-cis, and (e) 13-cis.

dynamics of excitation and relaxation, since each isomer is expected to keep the initial configuration immediately after excitation.

Before getting into details of time-resolved absorption spectroscopy on the present isomers, the previous results on the all-trans and the 9-cis isomers are summarized briefly by referring to ref 11: The all-trans isomer showed only one kind of triplet absorption at 440 nm, and this absorption grew continuously in the time domain of 40–100 ps (Figure 1 of ref 11). The λ_{max} of the absorption immediately ("0 ps") after excitation was apparently a little shifted toward the blue. The shift was ascribed to the facts that the arrival time of the interrogating pulse at the sample depends on the wavelength, and that blue light arrives a little later than red light. Thus, it is concluded that this isomer produces only one triplet species, whose concentration increases with time. The 9-cis isomer gave a unique absorption around 410 nm at "0 ps" after excitation (Figure 4 of ref 11). This absorption decayed with time (it remained until at least 40 ps after excitation), and a broad absorption centered around 440 nm built up. The broad absorption converged very slowly (within a few nanoseconds) into a sharp absorption, which was identical with that of the all-trans isomer.

Figure 2 shows the results for the present isomers, i.e., (a) 7-cis, (b) 11-cis, and (c) 13-cis. Figure 2a shows that the 7-cis isomer gives two absorption bands, one at 410 nm and the other at 440 nm. (The temporal distortion of the spectrum at "0 ps" after excitation should be taken into account in this case also; see also the set of spectra more than 40 ps after excitation.) The former decreases, while the latter increases with time. The λ_{max} of the latter corresponds to that of the triplet species of the all-trans isomer described above. Figure 2b shows that the 11-cis isomer also gives a unique absorption (<405 nm) immediately after excitation.[15] This absorption decays within less than 60 ps. Another absorption at 440 nm grows rapidly, and the spectral

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(15) At present, we cannot completely exclude the possibility that the strong peak in the spectrum recorded immediately after excitation ("0 ps") is due to the singlet excited state. However, in the case of retinylideneacetaldehyde, the $S_n \leftarrow S_1$ absorptions appear above 450 nm.[19]

(16) This type of sharpening of the $T_n \leftarrow T_1$ absorption, which reflects configurational relaxation in the triplet state, is observed in the case of the 9-cis isomer of retinylideneacetaldehyde, even after the subtraction of the $S_n \leftarrow S_1$ absorptions. The sharpening may indicate the presence of an intermediate, less stable triplet state between the primary "9-cis" and the stationary "all-trans" triplet species.

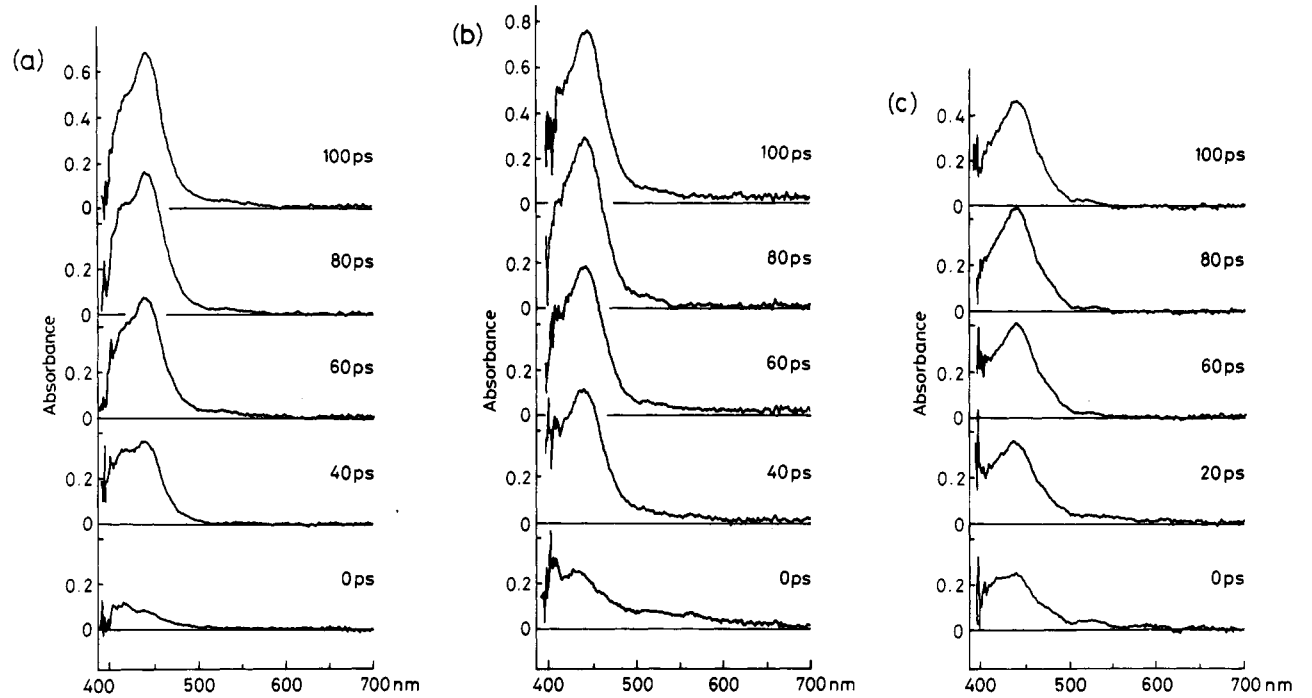

Figure 2. Transient absorption spectra recorded at "0"–100 ps after the excitation for the (a) 7-cis, (b) 11-cis, and (c) 13-cis isomers. ("0 ps" means that the pump and the probe pulses (both 20–25-ps duration) are temporally overlapped.)

pattern, recorded at 80 or 100 ps after excitation of this isomer, is already very similar to that of the all-trans isomer. (Spectral change in this isomer is much faster than that in the 7-cis isomer.) Figure 2c shows that the 13-cis isomer gives, immediately after excitation, an absorption with a shoulder in the region of shorter wavelengths. The shoulder may be ascribed, at least partly, to the temporal distortion of the spectrum; no sharp band was observed in the 400–450-nm region for this isomer. A sharpening of the broad absorption around 440 nm is also seen.

It is to be noted that the absorption at 440 nm, which is characteristic of the triplet species produced from the all-trans isomer, is clearly seen in the cases of the 7-cis and 11-cis isomers immediately after excitation ("0 ps"). (It is seen also in the case of the 9-cis isomer.[11]) This observation will be discussed in the last part of *Triplet Configurations and Isomerization Mechanisms.*

The Origin of the Transient Absorption: The Triplet State. Hochstrasser et al.[14] examined the transient absorption of the all-trans isomer in the 10-ps time scale (direct excitation at 353 nm in *n*-hexane). They detected two different kinds of transient absorption: an ordinary $T_n \leftarrow T_1$ absorption at 448 nm, which grew with a rate constant of 34 ± 5 ps, and an absorption at 640 nm ascribable to the equilibrated singlet excited state, which appeared instantaneously (<6 ps) and decayed in about 20 ps. However, we did not observe any absorption around 640 nm, even immediately after excitation.

We have estimated the rise time of the transient absorption of the all-trans isomer to be approximately 30 ps by the deconvolution method, taking into account the temporal characteristics of the exciting and interrogating pulses. This value is in good agreement with the value of 34 ± 5 ps cited above as the rise time of the $T_n \leftarrow T_1$ absorption. On the other hand, all the isomers examined here were practically nonfluorescent, which indicates that the excited singlet state is extremely short lived due to rapid intersystem crossing and/or to fast isomerization in the excited singlet state. These results strongly suggest that the transient absorptions observed here are due to the triplet state, not only for the all-trans isomer but also for the mono-cis isomers.[15]

Transformation among Triplet Species: Primary and Stationary Triplet. The spectral changes observed within 100 ps after excitation and the assignment of the absorption bands to the triplet state lead us to the concept of transformation between triplet species, i.e., from a primary to a stationary triplet species. The 7-cis and 11-cis isomers showed absorptions below 410 nm immediately after excitation, which we ascribe to "primary" triplet species. As these absorption decreased, the 440-nm absorption increased. Thus, it is concluded that these isomers produce primary triplet species of their own immediately after excitation, and that the primary triplet species transform into the stationary triplet species of the all-trans isomer. As described above, the all-trans isomer showed only one kind of triplet absorption at 440 nm. Therefore, this stationary triplet species can be produced soon after excitation (rise time 30 ps) when the all-trans isomer is used as starting material. The 13-cis isomer did not give any clear indication of a short-lived, primary triplet species. A broad band peaked at 440 nm remained 100 ps after excitation, a result that indicates the formation of the stationary triplet species of the all-trans isomer. Sharpening of the 440-nm band suggests the presence of a long-lived, primary triplet species inherent to the 13-cis isomer.

The comparison of spectra described above reveals that the cis isomers can be classified into two groups by the temporal characteristics of the triplet absorption: Unmethylated-cis isomers, i.e., the 7-cis and 11-cis isomers, show clear and fast transformation from a primary to a stationary triplet species. (The transformation of the 11-cis isomer is much faster.) Their stationary species are identical with the triplet species produced from the all-trans isomer. On the other hand, methylated-cis isomers give a very broad absorption during the relaxation into stationary triplet species. The stationary species produced from the 9-cis isomer is identical with the triplet species from the all-trans isomer, but the spectrum of the stationary species produced from the 13-cis isomer seems to have two different components (see below). The transformations of the methylated-cis isomers are slow and not simple.

Configurational Assignment of Primary Triplet Species. No information is available at present concerning the configurations of those primary and stationary triplet species. However, the simplest assumption would be that the primary triplet species is produced from each starting isomer, without any substantial configurational changes during the process of vibrational relaxation and intersystem crossing. In other words, we assume a set of potential minima in the triplet state, with different energies and barriers, each of which should be geometrically close to the potential minimum in the ground state. The identification of primary triplet species, each of which is characteristic of the starting isomer, supports the idea. From this viewpoint, we denote the isomeric configurations in the triplet state with quotation marks as "all-trans", "7-cis", "9-cis", "11-cis", and "13-cis"; these triplet configurations are produced from the all-trans, 7-cis, 9-cis, 11-cis,

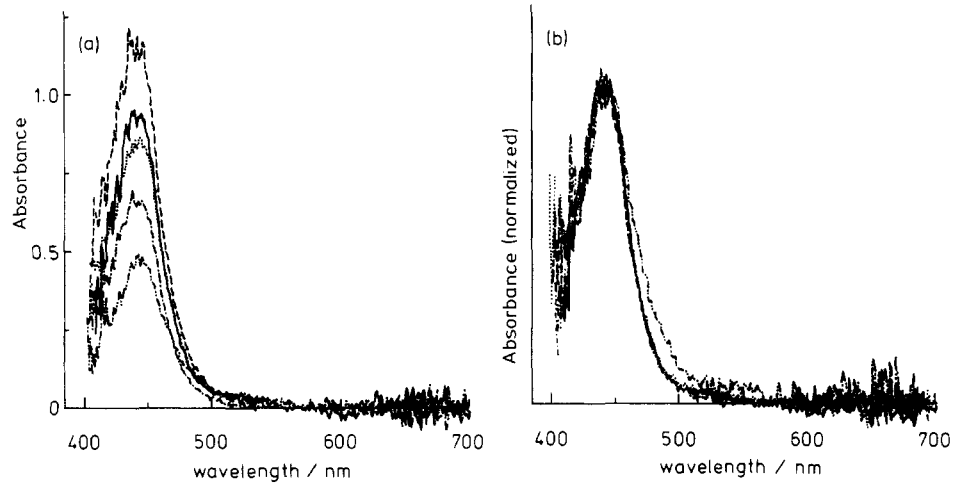

Figure 3. Transient absorption spectra recorded at 5 ns after excitation for the all-trans (—), 7-cis (···), 9-cis (- - -), 11-cis (·-·), and 13-cis (····) isomers. Spectra (a) before and (b) after normalization at λ_{max}.

and 13-cis ground-state configurations, respectively.

Stationary Triplet Species. Comparison of the Triplet Spectra Recorded 5 ns after Excitation and Previous Results of Resonance Raman Spectroscopy. Figure 3 compares transient absorption spectra recorded 5 ns after excitation of the all-trans, 7-cis, 9-cis, 11-cis, and 13-cis isomers; spectra both (a) before and (b) after normalization are shown. The agreement of the normalized spectra among all the isomers, except for 13-cis, confirms that only the stationary, "all-trans" triplet is present at this delay time. (The common absorption spectrum was designated pattern A.) The spectrum for the 13-cis isomer (doubly dotted broken line) is different and shows a tail on the longer wavelength side of the absorption, indicating the presence of another stationary triplet species. Comparison of the spectra on the shorter wavelength side is difficult, because the monitoring picosecond continuum is weak in this region.

A set of transient Raman spectra, recorded under conditions similar to those used for the present, time-resolved, electronic absorption measurements (direct excitation of the same set of isomers with the 355-nm pulses in *n*-hexane at room temperature), has provided more detailed information about the structures of the stationary triplet species.[3] The delay time between the pump and probe pulses was fixed at 20 ns. The all-trans, 7-cis, 9-cis, and 11-cis isomers gave an identical spectrum (we call this pattern A′ hereafter) within the limit of experimental error. The 13-cis isomer yielded a similar triplet spectrum, but it showed additional Raman lines that were absent from pattern A′. When the contribution of pattern A′ was subtracted, another type of Raman spectrum (pattern B′), which had the same number of Raman lines but with different frequencies and intensities, emerged. Both the pattern A′ and pattern B′ spectra could be explained in terms of one single triplet configuration; the number and the width of the Raman lines of each triplet species were not very different from those of each ground-state isomer. The pattern A′ and pattern B′ spectra were ascribed to "all-trans" and "13-cis" triplet species.[3,17]

The present results of transient absorption spectroscopy are consistent with the above results of the transient Raman spectroscopy. The pattern A absorption should definitely be assigned to one single triplet species, the "all-trans" triplet. The broader triplet absorption obtained from the 13-cis isomer should be ascribed to both the "all-trans" and the "13-cis" triplet. This assignment also supports the interpretation that the slow sharpening of the triplet absorption, observed in the case of the 13-cis isomer, is due to the transformation from the "13-cis" to the "all-trans" triplet.

HPLC Analyses of Photoisomerization Products and Configurational Assignment of Stationary Triplet Species. Figure 4 shows the results of the sensitized isomerization starting from the (a) all-trans, (b) 7-cis, (c) 9-cis, (d) 11-cis, and (e) 13-cis isomers. In these experiments, we focused on the primary product of isomerization. The results indicate the following: (1) All the mono-cis isomers produced only the all-trans isomer. The amount of the all-trans isomer produced were as follows: 13.5% for 7-cis; 9.3% for 9-cis; 18.0% for 11-cis; 9.3% for 13-cis. (The control experiment without the sensitizer (but with filters) caused practically no isomerization.) (2) The all-trans isomer produced only a very small amount of the 13-cis isomer (1.7%). (The amount of the product isomer in each case is expressed relative to the sum of the product and the starting isomers, which were present after exposure.)

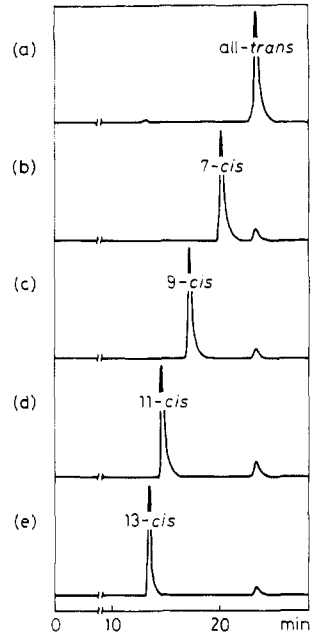

Figure 4. HPLC analyses of the products of benzil-sensitized photoisomerization for the (a) all-trans, (b) 7-cis, (c) 9-cis, (d) 11-cis, and (e) 13-cis isomers.

The one-way isomerization of the 7-cis, 9-cis, or 11-cis isomer into the all-trans isomer via the triplet state is very consistent with the one-way transformation of the triplet species, i.e., from "7-cis", "9-cis", or "11-cis" to "all-trans" (see Figure 4). Therefore, the overall configurational change between the initial and the final ground states through triplet formation, which is called "isomerization via triplet", is now definitely ascribed to the transformation between a pair of the triplet species. The one-way isomerization *into* the all-trans isomer supports the configurational assignment of the stationary "all-trans" triplet, which was originally introduced regarding the starting ground-state configuration.

(17) Koyama, Y. *Time-resolved Vibrational Spectroscopy*; Atkinson, G. H., Ed.; Gordon & Breach: New York, 1987; pp 15–40.

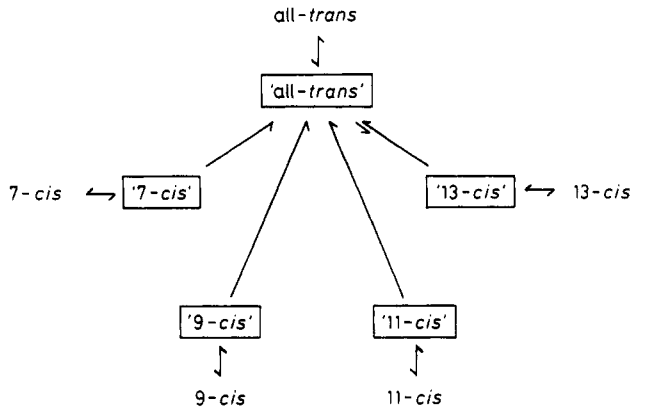

Figure 5. Diagram showing the isomerization through the configurational changes in the triplet state, when the molecules of retinal are excited from each ground-state configuration.

The all-trans isomer does isomerize into the 13-cis isomer via the triplet state, although the relative quantum yield of isomerization is low. This may indicate that the minimum of the "13-cis" triplet is almost as deep as the minimum of the "all-trans" triplet, and that it causes the long-lived "13-cis" triplet to stay more than 20 ns.

Triplet Configurations and Isomerization Mechanisms. In the present investigation, a piece of information about the mechanism of isomerization has been obtained by transient absorption spectroscopy on the triplet species and of the product analyses of benzil-sensitized photoisomerization. Before we get into detailed discussion, some qualifications are necessary. (1) We have to confine ourselves to the discussion of isomerization *via the triplet state*, since not the singlet but the triplet species were probed by the present absorption spectroscopy. (2) We discuss the triplet species produced at room temperature in a nonpolar solvent, *n*-hexane, since we have examined such a case here and since the triplet species produced can be dependent on the temperature and the polarity of the solvent. (3) The triplet species probed by absorption spectroscopy were produced by intersystem crossing from the S_1 state, while those used in the product analyses were produced by energy transfer from triplet benzil. We analyzed also the products of isomerization through direct excitation under the same conditions as those of the transient absorption measurements; the results were similar except for more pronounced isomerization around the C13=C14 bond of the isomers. We believe that the data for sensitized isomerization should be used in the discussion of isomerization via the triplet state.

Two alternative mechanisms of isomerization from a cis to the trans configuration in the triplet state are possible after intersystem crossing takes place from cis S_1* (the asterisk indicates species before vibrational relaxation). Mechanism a: Cis T_1* is produced from cis S_1*, cis T_1* relaxes vibrationally to the bottom of the cis potential minimum, and the resultant cis T_1 relaxes along the triplet potential surface (with a configurational change) to the bottom of the trans potential minimum (trans T_1). Mechanism b: Efficient partitioning of cis S_1* into cis T_1* and trans T_1* takes place, and then trans T_1* relaxes vibrationally to the bottom of the trans potential minimum (trans T_1).

In the present investigation, both the primary and the stationary triplet species have been detected, and transformation from "cis" primary species to the "trans" stationary species were actually seen by transient absorption spectroscopy. The present authors believe that this observation is the first strong evidence for mechanism a. The mechanism is summarized in Figure 5: The all-trans isomer generates only the "all-trans" triplet; no indication of configurational changes in the triplet state was seen. The 7-cis, 9-cis, and 11-cis isomers initially generate the "7-cis", "9-cis", and "11-cis" triplet species; all of them transform into the "all-trans" triplet. The rates of transformation in the triplet state are in the order, "11-cis > "7-cis" > "9-cis". The 13-cis isomer seems to produce a "13-cis" triplet, which does not relax completely into the "all-trans" triplet even 5 ns after excitation. The spectral changes accompanied by transformations suggest that mostly one-way transformations are present in the triplet state and that the potential minimum of the "all-trans" triplet is the deepest. The rates of transformations suggest that the relative heights of the potential barriers are in the order "11-cis" < "7-cis" < "9-cis" < "13-cis".

After relaxing along with the triplet potential surface into the deepest "all-trans" minimum, the molecules are supposed to relax, without any substantial configurational change, onto the ground-state potential surface. Assuming configurational similarity between the triplet and ground-state minima, the all-trans ground-state configuration is expected to be generated most frequently from those cis configurations. This prediction was shown to hold true by HPLC analysis of isomerization products.

Mechanism b has been proposed on the basis of the analysis of isomerization and transient absorption and Raman spectroscopy: The one-way isomerization of the 11-cis isomer into the all-trans isomer, a complete absence of the oxygen effect on the products of isomerization, and the identical triplet spectrum obtained from the 11-cis and the all-trans isomers led Rosenfeld et al.[4] to propose this mechanism ("vibronic mechanism"). Later, Hamaguchi et al.[3] detected, by resonance Raman spectroscopy, a single and common "all-trans" triplet species produced from all the mono-cis isomers except for 13-cis, and therefore, they also proposed mechanism b. They stated that efficient intersystem crossing and resultant partitioning into "cis" T_1* and "trans" T_1* are essential in understanding the isomerization mechanism. In both cases, the spectroscopic detection of a single, common triplet species in cis to trans isomerization played the key role in the proposition of this mechanism. The dependence of photoisomerization by direct excitation on the wavelength of excitation, as well as the dependence of sensitized photoisomerization on the donor energy,[18] was regarded as support for this mechanism.

The present investigation seems to give a new support to mechanism b also. In the set of spectra recorded immediately after excitation (the recordings denoted as "0 ps" in Figure 2, a and b), the transient absorption ascribable to the "all-trans" triplet was already seen together with cis triplets, although the exact estimation of its amount is difficult at the present stage because of the width (25 ps) of the pump and probe pulses. Measurements with shorter pulses are absolutely necessary to determine which mechanism is predominant in isomerization via the triplet state.

Registry No. all-*trans*-Retinal, 116-31-4; 7-*cis*-retinal, 24315-14-8; 9-*cis*-retinal, 514-85-2; 11-*cis*-retinal, 564-87-4; 13-*cis*-retinal, 472-86-6.

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